

Action of the pathogenesis-related protein PR10 from *Theobroma cacao* in triggering response mechanisms of *Moniliophthora perniciosa*

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Word Keys: Bi-dimensional gel, mass spectrometry, pathogenesis related-protein PR10, TcPR10 protein, Fungal disease

Cacao (*Theobroma cacao* L.) is an important commodity worldwide, but its production is limited by destructive diseases such as witches' broom, due to the fungus *Moniliophthora perniciosa*. The high recombination rate and genetic variability of this fungus promoted resistance breakdown of cacao and annihilated the efforts made by the Brazilian government to reduce the disease impact on plantations. According to the literature, pathogenesis related-proteins (PRs) play an important role in defense/resistance mechanisms of the plant submitted to various biotic and abiotic stresses. A cDNA clone encoding a PR10 (named TcPR10) was obtained from a cacao-*M. perniciosa* EST library. The corresponding recombinant protein (expressed in *Escherichia coli* BL21(DE₃)) presents a strong antifungal activity against *M. perniciosa*, as previously demonstrated. In this work, we developed a proteomic analysis of the fungus cultivated in the presence of TcPR10, using 2DE-MS/MS. *M. perniciosa* was grown in CPD 2% agar medium; after 15 days, the fungal hyphae were broken and were grown in presence of 3 µg/mL of TcPR10 for 1h. Then, the total proteins were extracted using the ADP method, followed by a simple cleaning using the method of SDS-dense and phenol. The quantification was made using a 2-D quantification kit. The proteins were extracted in triplicate and separated using a 12% bi-dimensional SDS-PAGE gel. The 2D map analysis showed approximately 300 "spots" per gel (control and one hour treatment) with differential protein expression pattern. The analysis using a mass spectrometry (nanoESI-Q-TOF) was made for the identification of the spots. We identified several proteins involved in fungal metabolism, carbohydrates/proteins metabolism, related proteins to growth and phytotoxins proteins. More spots have been identified to better understand the mechanism of fungi response to protein PR10.

Orgãos Financiadores: CNPq, CAPES